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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
09/710,058	11/10/2000	David Anderson	RIGL-011	4112

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EXAMINER	
LIU, SUE XU	

ART UNIT	PAPER NUMBER
1639	

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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	09/710,058	ANDERSON ET AL.	
	Examiner	Art Unit	
	SUE LIU	1639	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *13 March 2009*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-3,20 and 21* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-3, 20 and 21* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Continued Examination Under 37 CFR 1.114

1. A request for continued examination under 37 CFR 1.114 was filed in this application after a decision by the Board of Patent Appeals and Interferences, but before the filing of a Notice of Appeal to the Court of Appeals for the Federal Circuit or the commencement of a civil action. Since this application is eligible for continued examination under 37 CFR 1.114 and the fee set forth in 37 CFR 1.17(e) has been timely paid, the appeal has been withdrawn pursuant to 37 CFR 1.114 and prosecution in this application has been reopened pursuant to 37 CFR 1.114. Applicant's submission filed on 3/13/09 has been entered.

Claim Status

2. Claims 4-19 and 22 have been canceled as filed on 3/13/09.

Claims 1-3, 20 and 21 are currently pending and are being examined in this application.

Priority

3. This application claims priority to U.S. Provisional Patent Application Nos. 60/164,592, filed 11/10/1999, as previously acknowledged in the Office action mailed 3/22/06.

Claim Objection(s) / Rejection(s) Maintained

Claim Interpretation

4. The instant claim 1 recites "A retroviral vector comprising a polynucleotide encoding a green fluorescent protein (GFP) having the amino acid sequence of SEQ ID NO:2, wherein said retroviral vector is *capable of producing a stably transfected mammalian cell line comprising cells that contain said retroviral vector and that are fluorescent due to the expression of said GFP by said cells*."

To simplify, the said claim is drawn to a retroviral vector comprising a polynucleotide. The claim language also limits that the said polynucleotide encodes for a specific GFP, whose amino acid sequence comprises SEQ ID No:2. Thus, the scope of the instant claims encompasses any polynucleotide (DNA sequences) that encodes for the amino acid sequence of SEQ ID No:2. The specific amino acid sequence recited in the instant SEQ ID NO:2 is an exact match to the wild-type amino acid sequence for a *Renilla* (Sea Pansy) GFP.

The italic region (as indicated above) of the said claim is a recitation of intended use of the instant claimed "retroviral vector". Generally, recitation of intended uses is not afforded patentable weight. A recitation of the intended use of the claimed invention must result in a structural difference between the claimed invention and the prior art in order to patentably distinguish the claimed invention from the prior art. If the prior art structure is capable of performing the intended use, then it meets the claim.

In the instant case, as discussed below, the various combinations of the references would produce a retroviral vector comprising a polynucleotide (especially one with humanized codons)

encoding for the wildtype Renilla GFP that is capable of producing stably transfected mammalian cells that express GFP, without evidence to the contrary.

The following rejections are set forth in light of the above claim interpretations.

Claim Rejections - 35 USC § 103

5. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

6. This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

Anderson and Bryan

7. Claims 1, 3, 20 and 21 are rejected under 35 U.S.C. 103(a) as being obvious over **Anderson** et al (PNAS. Vol. 93: 8508-8511; 1996), in view of **Bryan** et al (US Patent 6,232,107; 2001; Filed 3/26/1999; priority date: 3/27/1998).

Anderson et al, throughout the publication, teach using retroviral vector expressing a GFP in mammalian cells. The reference teaches that retroviral gene transfer was used to stably incorporate the wildtype GFP (Aequorea) in a mammalian cell (see page 8509, left col., 1st para.

under RESULTS) and expressed in mammalian cells (NIH 3T3). The reference also teaches that FACS analysis of the retroviral vector comprising the wildtype GFP transduced cells revealed a single peak on a fluorescence histogram, and there was a two fold difference in fluorescence value between infected and uninfected cells (page 8509, left col., 1st para. under RESULTS). These would read on the fluorescence of the GFP can be detected by FACS since the fluorescence of the retroviral transduced cells in the reference was detected.

Anderson et al do not expressly teach that the cDNA for the GFP is humanized, and the specific GFP amino acid sequence (i.e. the Renilla wildtype GFP).

However, **Bryan** et al, throughout the publication, teaches a Renilla GFP (with specific amino acid sequence; i.e. amino acid sequence of SEQ ID NO:2) and its uses in various gene expression system. The reference discloses and claims the use (e.g. diagnostics and high throughput screening e.g. libraries) of nucleic acid molecules encoding green fluorescent proteins (e.g. bioluminscent) from the genus Renilla. Bryan further teaches protein Seq. Id. No. 16 which corresponds (e.g. has **100% sequence identity**) to "wild type" Renilla GFP of **Seq. Id. No.2**, as recited in the instant Claim 1. [see Reference Seq. Id 15 and attached Result 4 DATABASE Alignment search and Reference sequence Id. 16].

Bryan et al further teach host cells (e.g. present claims 1 and 3) including prokaryotic/eukaryotic (e.g. mammalian) which incorporate genetic constructs comprising a polynucleotide (e.g. cDNA) encoding a green fluorescent protein (GFP) having the amino acid sequence of SEQ ID No.2 (e.g. (sea pansy) wild-type *Renilla Mullerie* GFP) as well as the use of "human codon-optimized nucleic acid encoding a Renilla GFP" as in the present claim 20 (e.g. "The genes can be modified by substitution of codons optimized for expression in selected

host cells or hosts, such as humans and other mammals …"; See col. 5). In addition, the Bryan reference also teaches the DNA encoding for Renilla GFP are included in expression vectors for stable or transient expression (e.g. col.7, ll. 25+).

It is important to note that the Bryan reference, although teaching both (jellyfish) wild-type *Aequorea Victoria* GFP and (sea pansy) wild-type *Renilla Mullerie* GFP; the use of *Renilla* is strongly preferred due to the analytical problems present in the former. Particularly, *Aequorea* GFP possess two separate excitation peaks, whereas *Renilla* GFP has one excitation peak making it not ideal for use in analytical and diagonostic purposes:

"Consequently, (jellyfish) GFP mutants have been selected with the hope of **identifying mutants** that have *single excitation spectral peaks* shifted to the red (emphasis provided).

In fact **a stated purpose in constructing such mutants has been to attempt to make *A. Victoria* GFP more like the GFP from *Renilla*,** which has thus far not been cloned, but which has properties that make it far more ideal for use as an analytical tool. For many practical applications, the spectrum of Renilla GFP would be preferable to that of the Aequorea GFP, because *wavelength discrimination* between different fluorophores and detection of resonance energy transfer are easier if the component spectra are tall and narrow rather than low and broad". (emphasis provided; col.4, lines 54+).

See also '107, col. 3-5; col. 47-48;

Therefore, it would have been prima facie obvious for an ordinary skilled artisan to screen for generate a retroviral vector comprising a polynucleotide encoding a GFP with a specific amino acid sequence (e.g. wildtype Renilla GFP amino acid sequence) that is encoded

by a humanized cDNA for stable transfection of mammalian cells. Due to the advantages taught by Bryan et al that GFP allows rapid selection of retrovirally transduced mammalian cells and the motivation to use GFP in mammalian expression system as discussed supra (i.e. superior spectra property of Renilla GFP), a person of ordinary skill in the art would have been motivated at the time of the invention to construct a retroviral vector comprising a specific GFP (e.g. a humanized Renilla GFP) for using in a mammalian gene expression system. Since the construction of retroviral vector comprising various GFPs (including wildtype, mutant, or humanized) is known in the art (such as taught by Anderson et al), and the specific sequence of a Renilla GFP is known and is shown to be expressed in mammalian cells (as taught by Bryan et al), an ordinary skilled artisan would have been motivated to generate a retroviral vector comprising GFP having a specific amino acid sequence and a mammalian cell comprising the retroviral vector. In addition, because both the Anderson reference and Bryan reference teach methods of using stable expression vectors to express GFPs in various cells including mammalian cells, it would have been obvious to one skilled in the art to substitute one expression vector (or one type of GFP encoding nucleic acids) for the other to achieve the predictable result of making and using a retroviral vector comprising humanized GFP encoding nucleic acids.

An ordinary skilled artisan would have reasonable expectation of success of achieving such modifications since Anderson et al have demonstrated the success of generating retroviral vector comprising GFP used in mammalian cell expression.

Bryan and Aran

8. Claims 1-3 and 20 are rejected under 35 U.S.C. 103(a) as being unpatentable over **Bryan** et al. US Pat. No. 6,232,107 (5/01: filed 10/98 or earlier) with attached Result 4 DATABASE Alignment search and **Aran** et al. Cancer Gene Therapy, Vol. 5, No. 4 pages 195-206 (1998).

Bryan et al, disclose and claim the use (e.g. diagnostics and high throughput screening e.g. libraries) of nucleic acid molecules encoding green fluorescent proteins (e.g. bioluminscent) from the genus Renilla, including nucleotide reference Seq. ID No. 15 which is 98.4% (with best local similarity of 99.4%) homologous to elected Seq. ID No.1 (the nucleic acid sequence); differing by only one nucleotide (C vs. G). Bryan further teaches protein Seq. Id. No. 16 which corresponds (e.g. has **100% sequence identity**) to "wild type" Renilla GFP of **Seq. Id. No.2**, as recited in the instant Claim 1. [see Reference Seq. Id 15 and attached Result 4 DATABASE Alignment search and Reference sequence Id. 16].

Bryan et al further teach host cells (e.g. present claims 1 and 3) including prokaryotic/eukaryotic (e.g. mammalian) which incorporate genetic constructs comprising a polynucleotide (e.g. cDNA) encoding a green fluorescent protein (GFP) having the amino acid sequence of SEQ ID No.2 (e.g. (sea pansy) wild-type *Renilla Mullerie* GFP) as well as the use of "human codon-optimized nucleic acid encoding a Renilla GFP" as in the present claim 20 (e.g. "The genes can be modified by substitution of codons optimized for expression in selected host cells or hosts, such as humans and other mammals …"; See col. 5). In addition, the Bryan reference also teaches the DNA encoding for Renilla GFP are included in expression vectors for stable or transient expression (e.g. col.7, ll. 25+).

It is important to note that the Bryan reference, although teaching both (jellyfish) wild-type *Aequorea Victoria* GFP and (sea pansy) wild-type *Renilla Mullerie* GFP; the use of *Renilla* is strongly preferred due to the analytical problems present in the former. Particularly, *Aequorea* GFP possess two separate excitation peaks, whereas *Renilla* GFP has one excitation peak making it not ideal for use in analytical and diagonostic purposes:

"Consequently, (jellyfish) GFP mutants have been selected with the hope of **identifying mutants** that have *single excitation spectral peaks* shifted to the red (emphasis provided).

In fact **a stated purpose in constructing such mutants has been to attempt to make *A. Victoria* GFP more like the GFP from *Renilla*,** which has thus far not been cloned, but which has properties that make it far more ideal for use as an analytical tool. For many practical applications, the spectrum of Renilla GFP would be preferable to that of the Aequorea GFP, because *wavelength discrimination* between different fluorophores and detection of resonance energy transfer are easier if the component spectra are tall and narrow rather than low and broad". (emphasis provided; col.4, lines 54+).

See also '107, col. 3-5; col. 47-48;

The Bryan reference differs, if at all, from the presently claimed invention (e.g. see claims 1, 3 and 20) in failing to *explicitly teach* the use of a retrovirus as a vector.

However, in this regard, the Bryan reference teaches that a wide variety of multipurpose vectors suitable for the expression of heterologous proteins are known to those of skill in the art and are commercially available; with selection and use of such vehicles as being well within the skill of the artisan. In this regard, the Bryan vectors for use in mammalian hosts include

"**recombinant *virus***", as well as plasmid and phages e.g. the use of "**retroviral** long-terminal repeats and inducible promoters from other eukaryotic expression systems".. See e.g. col. 23 (especially bottom) to col. 24; col. 59-60 (emphasis provided). Accordingly, the Bryan reference taken alone provides motivation to select the use of retroviral vectors, especially for use in mammalian host cells.

Accordingly, it would have been obvious to one of ordinary skill in the art at the time of appellant's invention to select a retroviral vector for use in a cellular host (e.g. procarytoic or mammalian) with use of a genetic construct comprising a polynucleotide (e.g. cDNA) encoding a wild-type Renilla green fluorescent protein (GFP) or a fusion thereof with a reasonable expectation of success in light of the reference's ability to express Renilla GFP and in view of the benefits of using Renilla GFP (e.g. as compared to *A. Victoria* GFP.

To the extent that further motivation to select a retroviral vector is needed and to the extent that Bryan fails to teach the incorporation of an IRES site (e.g. in present claim 2) in its fusion constructs, the Aran reference is cited.

The **Aran et al**. reference teaches the favorable use of retroviral vectors, both in vitro and in vivo including an internal ribosome entry site (IRES) for fusion constructs preferably comprising optimized, humanized (e.g. see page 204, left column for benefits of humanizing) GFP (e.g. *Aequorea victoria*); since "[T]his vector allows rapid and specific identification of the expressed protein (e.g. MDR1 gene transfer) in living cells (e.g. mammalian cells) " (E.g. see Abstract and page 195, especially right column).

Accordingly, it would have been obvious to one of ordinary skill in the art at the time of applicants invention to utilize a retroviral vector as the Bryan "recombinant virus" vector with the use of an IRES for expressing humanized or non-humanized wild-type renilla GFP in the Bryan et al. reference in order to appreciate the benefits thereof; e.g. rapid and specific identification of the expressed protein.

Aran, Bryan and Zolutukhin

9. Claim 1-3 and 20 are rejected under 35 U.S.C. 103(a) as being unpatentable over **Aran** et al. (citation omitted), **Bryan** et al. (citation omitted) and, if necessary, further in view of Zolutukhin et al. US Pat. No. 5,874,304 (2/99: filed 1/96).

The above combined teaching of the Aran and Bryan references as described in the above obviousness rejection is hereby incorporated by reference in their entirety.

The combined reference teaching differs, if at all, from the presently claimed invention (e.g. claim 20) by failing to *explicitly* teach a human codon-optimized nucleic acid encoding a Renilla GFP (e.g. humanized GFP) in a retroviral vector.

However, **Zolutukhin et al** teach that utilizing human codon-optimized nucleic acid GFP in nucleic acid constructs (including fusion proteins; e.g. col. 4 corresponding to present claim 2 "first gene" terminology) serves to overcome prior art obstacles and is advantageous (e.g. improved expression in mammalian and human cells). These constructs are included in vectors (e.g. see col. 5, examples; particularly retroviral: see patent claims, especially claims 50 and 69) contained in cells (e.g. see patent claims 71-80) in which the constructs contain:

1. GFP (particularly Renilla: see col. 1, last paragraph; col. 14 and Table 1; and especially col. 16, lines 3-15: spectrum of Renilla ... preferable to that of Aequorea);

2. IRES elements (e.g. see '304 patent, col. 13, line 50; claims 50 and 62).

(See also col. 1-2).

Accordingly, one of ordinary skill in the art at the time of applicant's invention would have been motivated to utilize human codon-optimized nucleic acids expressing Renilla GFP in the genetic constructs (e.g. cells/vectors comprising renilla GFP/IRES elements) rendered obvious by the combined Aran et al and Bryan et al teaching in light of the advantages thereof imparted by such humanized sequences as taught by the Zolutukhin et al. reference.

Thus, it would have been prima facie obvious to one of ordinary skill at the time of appellant's invention to modify the cellular/vector genetic constructs taught by the Aran and Bryant reference to include human codon-optimized (e.g. humanized) nucleotides encoding renilla GFP in order to obtain the advantages thereof as taught by the Zolutukhin et al. reference.

Zolutukhin and Bryan

10. Claims 1-3, 20 and 21 are rejected under 35 U.S.C. 103(a) as being unpatentable over **Zolutukhin** et al. US Pat. No. 5,874,304 (2/99: filed 1/96) and **Bryan** et al. US Pat. No. 6,232,107 (5/01: filed 10/98 or earlier) with attached Result 4 DATABASE Alignment search., and if necessary in view of **Anderson** et al. (PNAS. Vol. 93: 8508-8511; 1996).

Zolutukhin et al teach that utilizing human codon-optimized nucleic acid GFP in nucleic acid constructs (including fusion proteins; e.g. col. 4 corresponding to present claim 2 "first

gene" terminology) serves to overcome prior art obstacles and is advantageous (e.g. improved expression in mammalian and human cells). These constructs are included in vectors (e.g. see col. 5, examples; particularly retroviral: see patent claims, especially claims 50 and 69) contained in cells (e.g. see patent claims 71-80) in which the constructs contain:

1. GFP (particularly Renilla: see col. 1, last paragraph; col. 14 and Table 1; and especially col. 16, lines 3-15: spectrum of Renilla ... preferable to that of Aequorea);

2. IRES elements (e.g. see '304 patent, col. 13, line 50; claims 50 and 62).

(See also col. 1-2).

It is noteworthy that Zolutukhin teaches that (sea pansy) Renilla GFP is more preferable as a reporter than Aequorea GFP since Aequorea has two absorbance peaks whereas Renilla GFP has a single absorbance peak at 498 accordingly:

For many practical applications, the spectrum of Renilla GFP would be preferable to that of Aequorea because wavelength discrimination between different fluorophores and detection of resonance energy transfer are easier when the component spectra are tall and narrow rather than low and broad." Accordingly, Zolutukhin (like the Bryan reference) teaches mutation of Aequorea toward obtaining a single peak (e.g. like Renilla) is desired. (Zolutukhin, col. 16, especially lines 10+).

Although the Zolutukhin et al. reference teaches nucleic acid which employ the preferential use of Renilla GFP, the Zolutukhin reference differs from the presently claimed invention by failing to explicitly teach the use of a *Renilla* GFP gene sequence which encodes wild type Renilla GFP corresponding to SEQ ID NO.2.

Bryan et al disclose and claim the use (e.g. diagnostics and high throughput screening e.g. libraries) of nucleic acid molecules encoding green fluorescent proteins (e.g. bioluminscent) from the genus Renilla, including nucleotide reference Seq. ID No. 15 which is 98.4% (with best local similarity of 99.4%) homologous to elected seq. ID 1; differing by only one nucleotide (C vs. G). Bryan further teaches protein SEQ ID No. 16 which corresponds (e.g. has **100% sequence identity**) to "wild type" Renilla GFP of **SEQ ID NO.2**, as presently claimed [see the attached Result 4 DATABASE Alignment for comparison between the instant SEQ ID No.1 and the reference's SEQ ID NO. 15].

Bryan et al further teach host cells (e.g. present claims 1 and 3) including prokaryotic/eukaryotic (e.g. mammalian) which incorporate genetic constructs comprising a polynucleotide (e.g. cDNA) encoding a green fluorescent protein (GFP) having the amino acid sequence of SEQ ID 2 (e.g. (sea pansy) wild-type *Renilla Mullerie* GFP) as well as the use of "human codon-optimized nucleic acid encoding a Renilla GFP" as in present claim 20 (e.g. "The genes can be modified by substitution of codons optimized for expression in selected host cells or hosts, such as humans and other mammals …" See col. 5). The reference further teaches the use of a fusion partner (e.g. a targeting agent as a first gene) in its genetic fusion constructs as in present claims 2 and 3. See e.g. col. 8; col. 11-15; col. 24. In addition, the Bryan reference also teaches the DNA encoding for Renilla GFP are included in expression vectors for stable or transient expression (e.g. col.7, ll. 25+).

Bryan et al teach the use of the bioluminescent green fluorescent proteins in cellular assays (e.g. live cells, including mammalian) and in high throughput screening systems (e.g. employing libraries) (e.g. see col. 2-3; 14).

It is important to note that the Bryan et al. reference, although teaching both (jellyfish) wild-type *Aequorea Victoria* GFP and (sea pansy) wild-type *Renilla Mullerie* GFP; the use of *Renilla* is strongly preferred due to the analytical problems present in the former. Particularly, *Aequorea* GFP possess two separate excitation peaks, whereas *Renilla* GFP has one excitation peak making it not ideal for use in analytical and diagonostic purposes:

"Consequently, (jellyfish) GFP mutants have been selected with the hope of **identifying mutants** that have *single excitation spectral peaks* shifted to the red (emphasis provided).

In fact **a stated purpose in constructing such mutants has been to attempt to make *A. Victoria* GFP more like the GFP from *Renilla*,** which has thus far not been cloned, but which has properties that make it far more ideal for use as an analytical tool. For many practical applications, the spectrum of Renilla GFP would be preferable to that of the Aequorea GFP, because *wavelength discrimination* between different fluorophores and detection of resonance energy transfer are easier if the component spectra are tall and narrow rather than low and broad". (emphasis provided; col.4, lines 54+).

See also '107, col. 3-5; col. 47-48;

In addition, **Anderson** et al, throughout the publication, teach using retroviral vector expressing a GFP in mammalian cells, as discussed supra. The discussion regarding the Anderson reference above is hereby incorporated by reference in its entirety.

Thus, it would have been obvious to one of ordinary skill in the art at the time of applicant's invention to utilize the Bryan polynucleotide Renilla green fluorescent protein

(including seq. Id 15) in the Zolutukhin reference genetic constructs for the stable expression of GFP in mammalian cells as taught by Zolutukhin and/or Anderson since:

a. BOTH Zolutukin and Bryan teach the preferential use of Renilla GFP thus motivating the selection of the Bryan Renilla GFP obvious to one of ordinary skill in the art;

b. Zulutukin teaches humanized polynucleotides encoding for GFP improves the expression vector especially for stable expression in mammalian (or human) cell lines; and/or

c. one of ordinary skill in the art would have been motivated to select the Bryan reference Renilla sequences for purposes of performing screening assays (e.g. high throughput library screens) in order to obtain the benefits of the renilla protein in such assays as taught by the Bryan reference.

Bierhuizen and Bryan

11. Claims 1, 3, 20 and 21 are rejected under 35 U.S.C. 103(a) as being obvious over **Bierhuizen** et al (Biochemical and Biophysical Research Communications. Vol. 234: 371-375; 1997), in view of **Bryan** et al (US Patent 6,232,107; 2001; Filed 3/26/1999; priority date: 3/27/1998).

Bierhuizen et al teach using retroviral vector to transfer Green Fluorescent Protein (GFP) into mammalian cells (see Abstract of the reference). The reference teaches both wildtype and mutant (in term of amino acid sequence) of Aequorea Victoria GFP were stably expressed in mammalian cells (see Abstract; Figure 1; page 373, left col., 1st paragraph; and page 374, left col., 1st paragraph). The reference also teaches that retroviral vectors comprising various GFP

constructs were generated (See page 373, left col., 1st paragraph), which would read on a retroviral vector comprising a polynucleotide encoding a green fluorescent protein. In addition, the reference teaches that FACS was used to analyze GFP expression (See Figure 1 and caption). The reference further teaches that humanized (meaning replacing Aequorea Victoria codons with human codons in the coding DNA sequence) GFP can achieve higher expression in mammalian cells (Page 371, right col., last lines of 2nd paragraph). Furthermore, the reference teaches that the purpose of the study was to evaluate the potential applicability of GFP expression as a marker for the rapid selection of retrovirally transduced mammalian cells (See page 374, left col., 1st line). The study of the reference conclusively teaches that the data showed that all variants (including the wildtype GFP) allow for flow cytometric detection (FACS) of stable GFP expression in mammalian cells and that he expression can be transferred by the MFG retroviral vector (See page 374, left col., 1st paragraph, last lines).

Bierhuizen et al do not expressly teach the Renilla GFP with the specific amino acid sequence recited in SEQ ID No 2.

However, **Bryan** et al teach the use of Renilla GFP as described in the rejection under "Bryan and Aran", and is incorporated by reference to its entirety as set forth below. The reference teaches use (e.g. diagnostics and high throughput screening e.g. libraries) of nucleic acid molecules encoding green fluorescent proteins (e.g. bioluminscent) from the genus Renilla, including Seq. Id. No. 16 which corresponds (e.g. has 100% sequence identity) to "wild type" Renilla GFP of Seq. Id. 2, as presently claimed.

Bryan et al further teach host cells (e.g. present claims 1 and 3) including prokaryotic/eukaryotic (e.g. mammalian) which incorporate genetic constructs comprising a

polynucleotide (e.g. cDNA) encoding a green fluorescent protein (GFP) having the amino acid sequence of SEQ ID 2 (e.g. (sea pansy) wild-type Renilla Mullerie GFP) as well as the use of "human codon-optimized nucleic acid encoding a Renilla GFP" as in present claim 20 (e.g. "The genes can be modified by substitution of codons optimized for expression in selected host cells or hosts, such as humans and other mammals …" . See col. 5). The reference further teaches the use of a fusion partner (e.g. a targeting agent as a first gene) in its genetic fusion constructs as in present claims 2 and 3. See e.g. col. 8; col. 11-15; col. 24. Additionally, Bryan et al teach the use of the bioluminescent green fluorescent proteins in cellular assays (e.g. live cells, including mammalian) and in high throughput screening systems (e.g. employing libraries) (e.g. see col. 2-3; 14). In addition, the Bryan reference also teaches the DNA encoding for Renilla GFP are included in expression vectors for stable or transient expression (e.g. col.7, ll. 25+).

It is important to note that the Bryan reference, although teaching both (jellyfish) wild-type *Aequorea Victoria* GFP and (sea pansy) wild-type *Renilla Mullerie* GFP; the use of *Renilla* is strongly preferred due to the analytical problems present in the former. Particularly, *Aequorea* GFP possess two separate excitation peaks, whereas *Renilla* GFP has one excitation peak making it not ideal for use in analytical and diagonostic purposes:

"Consequently, (jellyfish) GFP mutants have been selected with the hope of **identifying mutants** that have *single excitation spectral peaks* shifted to the red (emphasis provided).

In fact **a stated purpose in constructing such mutants has been to attempt to make *A. Victoria* GFP more like the GFP from *Renilla*,** which has thus far not been cloned, but which has properties that make it far more ideal for use as an analytical tool. For many practical

applications, the spectrum of Renilla GFP would be preferable to that of the Aequorea GFP, because *wavelength discrimination* between different fluorophores and detection of resonance energy transfer are easier if the component spectra are tall and narrow rather than low and broad". (emphasis provided; col.4, lines 54+).

See also '107, col. 3-5; col. 47-48;

The Bryan reference differs, if at all, from the presently claimed invention (e.g. see claims 1, 3 and 20) in failing to *explicitly teach* the use of a retrovirus as a vector.

However, in this regard, the Bryan reference teaches that a wide variety of multipurpose vectors suitable for the expression of heterologous proteins are known to those of skill in the art and are commercially available; with selection and use of such vehicles as being well within the skill of the artisan. In this regard, the Bryan vectors for use in mammalian hosts include "**recombinant *virus***", as well as plasmid and phages e.g. the use of "**retroviral** long-terminal repeats and inducible promoters from other eukaryotic expression systems". See e.g. col. 23 (especially bottom) to col. 24; col. 59-60 (emphasis provided). Accordingly, the Bryan reference taken alone provides motivation to select the use of retroviral vectors, especially for use in mammalian host cells.

Accordingly, it would have been obvious to one of ordinary skill in the art at the time of applicant's invention to select a retroviral vector for use in a cellular host (e.g. prokaryotic or mammalian) with use of a genetic construct comprising a polynucleotide (e.g. cDNA) encoding a wild-type Renilla green fluorescent protein (GFP) or a fusion thereof with a reasonable expectation of success in light of the reference's ability to express Renilla GFP and in view of the benefits of using Renilla GFP (e.g. as compared to *A. Victoria* GFP).

Therefore, it would have been prima facie obvious for an ordinary skilled artisan to screen for generate a retroviral vector comprising a polynucleotide encoding a GFP with a specific amino acid sequence (e.g. wildtype Renilla GFP amino acid sequence). Due to the advantages taught by both Bierhuizen et al and Bryan et al that GFP allows rapid selection of retrovirally transduced mammalian cells, a person of ordinary skill in the art would have been motivated at the time of the invention to construct a retroviral vector comprising a specific GFP (e.g. Renilla GFP) for using in a mammalian gene expression system. Since the construction of retroviral vector comprising various GFPs (including wildtype, mutant, or humanized) is known in the art (such as taught by Bierhuizen et al), and the specific sequence of a Renilla GFP is known and expressable in mammalian cells (as taught by Bryan et al), an ordinary skilled artisan would have been motivated to generate a retroviral vector comprising GFP having a specific amino acid sequence and a mammalian cell comprising the retroviral vector. An ordinary skilled artisan would have reasonable expectation of success of achieving such modifications since Bierhuizen et al have demonstrated the success of generating retroviral vector comprising GFP used in mammalian cell expression.

In conclusion, the invention of the instant claims would have been prima facie obvious over Bierhuizen et al, in view of Bryan et al to one of ordinary skill in the art without evidence to the contrary.

Bierhuizen, Bryan and Aran

12. Claims 1-3, 20 and 21 are rejected under 35 U.S.C. 103(a) as being obvious over **Bierhuizen** et al (Biochemical and Biophysical Research Communications. Vol. 234: 371-375;

1997), in view of **Bryan** et al (US Patent 6,232,107; 2001; Filed 3/26/1999; priority date: 3/27/1998) and further in view of **Aran** et al (Cancer Gene Therapy. Vol. 5: 195-206; 1998).

Bierhuizen et al teach a GFP retroviral vector and mammalian gene expression system as described supra under "Bierhuizen and Bryan", and is hereby incorporated by reference in its entirety.

Bryan et al teach a Renilla GFP and its uses in various gene expression system as described supra under "Bryan and Aran", and is hereby incorporated by reference in its entirety.

Both of the references do not expressly teach the expression vector comprises IRES.

However, **Aran** et al teach a retroviral vector comprising an IRES element (See Page 197, left col., 2nd paragraph). The reference also teaches that the retroviral vector comprises a GFP and is used to transducer a mammalian cell (page 197, left col., 2nd and last paragraphs). The reference further teaches the advantage of including IRES element such as the element allows efficient cap-independent translation of the downstream gene.

Therefore, it would have been prima facie obvious for an ordinary skilled artisan to screen for generate a retroviral vector comprising a polynucleotide encoding a GFP with a specific amino acid sequence (e.g. wildtype Renilla GFP amino acid sequence) and an IRES element. Due to the advantages taught by Aran et al that the inclusion of an IRES element in a retroviral vector expression system would facilitate gene expression, an ordinary skilled artisan would be motivated at the time of the invention to generate a retroviral GFP expression vector comprising an IRES element. In addition, the inclusion of an IRES element in a eukaryotic gene expression system is well known in art such as taught by Aran et al. An ordinary skilled artisan would have reasonable expectation of success of achieving such modifications since Aran et al

have demonstrated the success of generating retroviral vector comprising GFP and IRES used in mammalian cell expression system.

In conclusion, the invention of the instant claims would have been prima facie obvious over Bierhuizen et al, in view of Bryan et al and further in view of Aran et al to one of ordinary skill in the art without evidence to the contrary.

Discussion and Answer to Argument

13. Applicant's arguments have been fully considered but they are not persuasive for the following reasons (in addition to reasons of record). Each point of applicant's traversal is addressed below (applicant's arguments are in italic):

Applicants assert the claim amendments (as filed on 3/13/09) would overcome all of the outstanding claim rejections. (Reply, pp 4+).

Applicants are respectfully directed to the above modified rejections for detailed discussion on how the combinations of the references render the instant claimed invention obvious. Applicants are also respectfully directed to the previously mailed Examiner Answer for answer to arguments presented in the Appeal Brief.

In addition, applicants are also respectfully directed to the Board decision regarding various issues raised in the Appeal Brief.

Briefly, the board decision is summarized below:

1.) In regard to the "teach away" argument using the Aran, Hanzano, Levy, Cheng and Anderson references, the Board found applicant's arguments not persuasive (decision, pp.9+). The decision states "the preponderance of the evidence on this record support a conclusion that a

person of ordinary skill in the art at the time this invention was made would have reasonably expected that a retrovial vector expressing Renilla GFP would produced detectable fluorescence in the host cell." (Decision, p.10).

2.) Bryan and Aran Rejection: The board states "Therefore, the prior art recognized that in order to obtain fluorescence of A. Victoria GFP in a host cell, a person of ordinary skill in the art modifies the A. Victoria GFP to make it more like Renilla GFP. For this reason, a person of ordinary skill in the art at the time of Appellants' claimed invention would have reasonably expected that a retrovial vector expressing Renilla GFP would have produced detectable fluorescence in a host cell. "In determining whether obviousness is established by combining the teachings of the prior art, the test is what the combined teachings of the references would have suggested to those of ordinary skill in the art." In re GPAC Inc., 57 F.3d at 1581 (internal quotations omitted). Accordingly, we are not persuaded by Appellants' contention to the contrary." (Decision, p.12; emphasis added).

3.) Aran, Bryan and Zolotukhin: The board was not persuaded by applicant's argument for the same reason as discussed above in 2.).

4.) Zolutokhin and Bryan: The board was not persuaded by applicant's argument for the same reason as discussed above in 2.).

5.) Bierhuizen and Bryan: The board was not persuaded by applicant's argument for the same reason as discussed above in 2.).

6.) Bierhuizen, Bryan and Aran: The board was not persuaded by applicant's argument for the same reason as discussed above in 2.).

7.) Anderson and Bryan: The board was not persuaded by applicant's argument for the same reason as discussed above in 2.).

New Claim Objection(s) / Rejection(s)

Claim Rejections - 35 USC § 112

14. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

15. Claim 21 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 21 recites "The retroviral vector… wherein fluorescent of said GFP can be detected by fluorescence-activated cell sorting (FACS)," which is unclear. The instant claim 21 seems to recite using FACS to sort/detect the "retroviral vector", which the said retroviral vector is not a "cell".

Conclusion

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Sue Liu whose telephone number is 571-272-5539. The examiner can normally be reached on M-F 9am-3pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Christopher Low can be reached at 571-272-0951. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

/Sue Liu/
Primary Examiner, AU 1639
5/18/09